EFEKTIVITAS THIACLOPRID, DIAFENTHIURON, DAN PETROLEUM SPRAY OIL UNTUK PENGELOLAAN *Bemisia tabaci* DAN PENYAKIT KUNING

The Effectiveness of Thiacloprid, Diafenthiuron, and Petroleum Spray Oil for Management of Bemisia tabaci *and Yellow Curl disease*

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INTISARI

B. tabaci (Homoptera; *Aleyrodidae*) merupakan hama penting karena dapat menimbulkan kerusakan langsung pada tanaman cabe dan berperan sebagai vector penyakit virus. Penelitian ini dilaksanakan dengan tujuan untuk mengetahui efektifitas tiga jenis insektisida dalam mengendalikan populasi *B. tabaci* dan penyakit daun kuning. Penelitian dilaksanakan di Muntilan, Jawa Tengah dengan rancangan acak kelompok lengkap terdiri atas empat perlakuan (thiacloprid, PSO, diafenthiuron, dan kontrol) masing-masing dengan empat ulangan. Insektisida diaplikasikan sesuai dengan dosis anjuran dengan interval satu minggu. Penyemprotan pertama kali dilaksanakan pada umur 7 hari setelah tanam dan diakhiri pada unur 98 hari sesudah tanam.

Aplikasi insektisida setiap minggu mampu menekan populasi *B. tabaci* pada saat terjadi puncak kepadatan populasi. Diafenthiuron mampu menekan populasi dewasa dan nimfa, thiacloprid mampu menekan populasi dewasa, dan PSO mampu menekan populasi nimfa *B. tabaci*. Namun demikian, penurunan populasi *B. tabaci* tidak menghambat perkembangan penyakit di lapangan. Deteksi molecular menunjukkan bahwa penyakit keriting kuning yang menyerang cabe disebabkan oleh begomovirus.

Kata-kata kunci: *Bemisia tabaci* – thiacloprid – PSO – diafenthiuron

INTRODUCTION

Chili (*Capsicum annuum* L.) is one of important tropical vegetables in term of its value and production area. The chili production worldwide is limited by several factors, among them are pests and diseases. One among the important pests is the whitefly, *Bemisia tabaci* (Gennadius). The principal reason for its status as a vegetables pest is that it vectors a number of viral pathogens (Byrne and Draeger, 1989), such as

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...rus, potyvirus, nepovirus, luteovirus, and a DNA-containing rod-shaped virus (Duffus, 1996 *cit.* Oliveira *et al.*, 2001). At high population levels, feeding is believed to cause direct damage to cotton and various vegetables crops (Byrne and Draeger, 1989).

Recently, *B. tabaci* becomes an important pest of chili in Indonesia because it transmits viruses that causes a disease with appearance of leaves damage such as curling, cramping, twisting, chlorosis, and dwarfing (Aidawati *et al.*, 2001; Sumardiyono *et al.*, 2003). In 2002, most of chili plants in Sleman (The Special Teritory of Jogjakarta) and Magelang (Central Java) was attacked by this disease with the disease intensity ranged from 20 to 100% for bell pepper (*C. annuum*), while for hot chili (*C. frustescens*) was 50 to 100% (Sumardiyono *et al.*, 2003).

The use of insecticides was the primary strategy currently employed to control *B. tabaci* (Polston and Anderson, 1997; Palumbo *et al.*, 2001b). Conventional insecticides commonly used during the past few years were organophosphate, carbamates, and pyrethroids (Forer 1990 *cit.* Horowitz and Ishaaya, 1992; Brazzle *et al.*, 1997; Nakhla and Maxwell, 1998) and their effectiveness was often dependent on sprays coverage and deposition (Sharaf 1986 *cit.* Palumbo *et al.*, 2001a). Ineffectiveness of insecticides applications was caused by rapid rate of reproduction and extensive host range of *B. tabaci* (Naranjo and Flint, 1994; Toscano *et al.*, 1997). A few new classes of insecticides (neonicotinoids, insect growth regulator (IGR)) have been developed recently and some have been reported effectively controlling *B. tabaci* (Palumbo *et al.*, 2001b).

In one of the chili production area, Muntilan, growers have used insecticides intensively but the disease has continue to significantly reduce the yield. Therefore, chili study was intended to evaluate the effectiveness of the most currently used insecticide, diafenthiuron, in the area, and two other insecticides with novel mode of action thiacloprid and petroleum spray oil (PSO).

Thiacloprid, second generation of nicotinoids, has a mode of action similar to other nicotinoids that attack the nervous system, causing irreversible blockage of postsynaptic nicotinergic acetylcholine receptors (Liu and Casidi 1993 *cit* Palumbo *et al.*, 2001b). PSO is thought to act directly on insects by blocking the spiracles and causing suffocation. An additional effect brought about by coating olfactory receptor organs may cause interference with host location (Simons 1982 *cit* Stansly *et al.*, 2000). Diafenthiuron is an insecticide that has been mostly used in the Magelang. It directly affects insect respiration through the inhibition of oxidative phosphorylation and disruption of mitochondrial ATP synthesis (Kayser 1993 *cit.* Palumbo *et al.*, 2001a).

MATERIALS AND METHODS

Experimental design

Thiacloprid 240 g AI/L (Calypso 240 EC, Bayer CropScience, Indonesia) and diafentiuron 500 g AI/L (Pegassus 250 EC, Syngenta, Indonesia) were obtained from local store, while PSO was obtained from SK Corporation Korea under University of Western Sydney authority.

The studies were conducted in Muntilan, The Province of Central Java, starting from July to December 2003. The field was set up in completely randomized block design (CRBD) consisting of four treatments: thiacloprid (0.5 ml/L), diafenthiuron (2 ml/L), PSO (0.5%), and unsprayed control and each treatment was replicated four times. The insecticide applications were carried out using a backpack sprayer with a spray volume of 750 l/ha. The first spray was conducted at the first week after transplanting, and the following sprays were done with an interval of seven days until a week before harvest (98 days after transplanting, DAT).

Cultural practices

In the nursery, chili seedlings (*C. annuum*) var. OR 369 (Oriental Seed, Indonesia) was sprayed with imidacloprid 200 g AI/L (Confidor 200 SL, Bayer CropScience, Indonesia). Seedlings were transplanted after they had four leaves. Before transplantation, holes (10 cm depth, 6 cm diameter) were prepared for planting the seedlings. These holes were filled with organic soil and sprayed with betasiflutrin 25 g AI/L (Buldok 25EC, Bayer CropScience, Indonesia) to protect the early planted seedlings from the soilworm, *Agrotis* spp.

The beds (1.2 m width and 12.5 m length) were covered with plastic, except holes for the seedlings. Seedlings were transplanted individually in beds consisting of two rows. The distance among rows was 60 cm with 50 cm apart within each row. Blocks were bordered by one bed planted with bean (*Vigna sinensis*). Beds in each block were spaced 1 m.

Community local cultural practices were used in the preparation and maintenance of experimental crops. The crops were fertilized three times during the growing season. The first fertilizer, compound fertilizer (Planta, 4 g/hole, Saraswati Anugerah Makmur, Sidoarjo, North Java), was applied a day before planting. A month after planting, the NP fertilizers (DAP) were applied at a rate of 5.2 g/L by spraying the soil around the plants. A week before the first harvest (98 DAT), plant growth regulator and compound fertilizer (Neo Kristalon, Saprotan Utama, Semarang, Central Java) mixed with fungicide difenokonazol 250 g/L (Score 250 C, Novartis, Indonesia) were sprayed on the plants, at rates of 1 g/L, 1 ml/L, and 7 ml/L, respectively. After fertilization, all plots were irrigated until the water reached the level of 10 cm below the ground surface for each bed. The land was soaked for 24 hours.

Observations

Efficacy of the insecticides. *Population density*. Relative efficacy of insecticides treatments in suppressing *B. tabaci* population was measured by determining adults and nymph density in the treated and untreated plots. The density of *B. tabaci* was counted by choosing randomly 20 plants per plot. Observations on adult density were done by counting *B. tabaci* at all fully expanded leaf of the 3rd and 4th node from the top of the plants, while the nymphs on the 5th to 7th terminal leaflet because of continued growth of the plant after oviposition (USDA, 2003). Observations were carried out by gently inverting the leaf.

Disease Incidence. Plants were monitored weekly for symptoms of virus disease. All the plants in the plots that showed virus disease symptoms were marked and observed visually to see the severity of the disease.

Yield. Yields were evaluated by harvesting the fruits from 20 plants selected randomly from each treatment. In addition, the total yield was also recorded by weighting total chili fruits from each plot. Harvest was conducted once a week, and the weight of chili every harvest time was recorded for each treatment.

Confirmation of the disease. Chili leaves that showed disease symptoms were used for molecular detection of the viral pathogen. DNA for Polymerase Chain Reaction (PCR) was prepared from field samples using CTAB method.

The PCR were carried out using a set of universal primers of begomovirus (Briddon and Markham, 1994) to amplify 2.8 kbp whole genom. Reaction was carried out in thermocycler as follows: 3 min at 96^{0}C (one cycled); followed by 30 s at 96^{0}C, 30 s at 50^{0}C, and 3 min at 72^{0}C (thirty five cycles); with the extension cycle of 5 min at 72^{0}C. PCR product was analysed by electrophoresis in 1% agarose gel containing 1x TBE buffer 89 mM Tris-borate, 2mM Na_2EDTA ph 8). The product was visualized under UV trans illuminator.

Analysis

The relative efficacy of tested insecticides in controlling *B. tabaci* population in the field was measured based on data on the number of *B. tabaci* and yield. Data were transformed using $\sqrt{\bar{x}}$ transformation to stabilize variances. Untransformed means were presented in tables and figures. Means were compared among insecticides treatments using ANOVA, and Fisher's protected LSD at α=5% was applied when ANOVA test indicated significance.

RESULTS AND DISCUSSION

Efficacy of insecticides

Population densities. The population densities of *B. tabaci* as a result of insecticides treatment were low at the beginning of the season, but later in the season when chili fruits mature (73-94 DAT) the population reached the peak. The population started to decrease as the fruits were harvested (105 DAT) or the crops deteriorated. Only at the peak of population (73, 80, 87, 94, 101 DAT) significant differences were observed. *B. tabaci* caught from the thiacloprid and diafenthiuron plots were significantly lower than that of the control plots (Fig. 1).

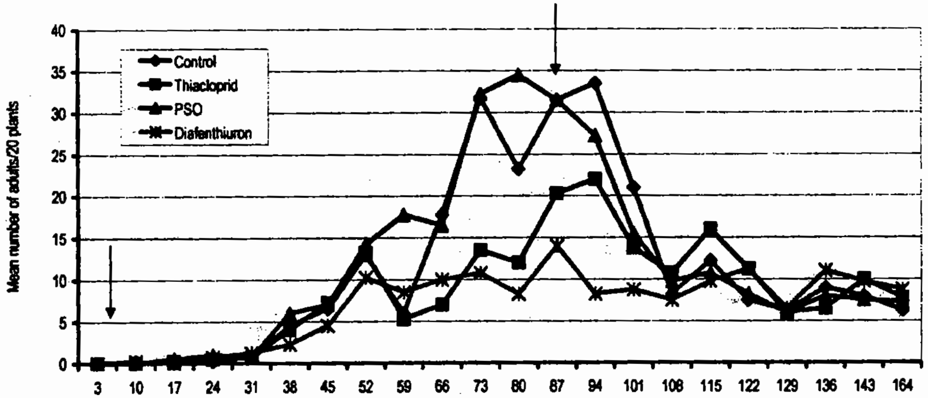

Fig. 1. Population densities of adults of *Bemisia tabaci* in the field plots treated with three different insecticides. The first arrow showed the first insecticides application, while the second arrow represented the last insecticides application. Insecticides were applied weekly. Observations were conducted directly by leafturn method.

The trend of nymph population was similar to that of the adult. Population started to decrease as the crops deteriorated. Furthermore, the densities of the nymph in the PSO and diafenthiuron plots were significantly lower than those in the control plots at 73, 80, 87, 94, 101, 108, 115, 122, 129, 136, 143 DAT. However, the nymph density in the plots treated with thiacloprid had similar number with that in the control plots (87, 94, 108, 129, 136 DAT; Fig. 2).

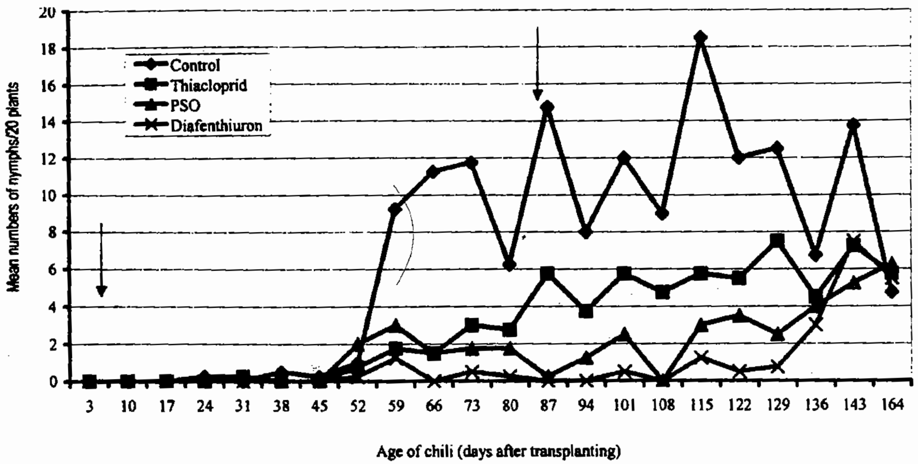

Fig. 2. Population densities of nymphs of *Bemisia tabaci* in the field plots treated with three different insecticides. The first arrow showed the first insecticides application, while the second arrow represented the last insecticides application. Insecticides were applied weekly. Observations were conducted directly by leafturn method.

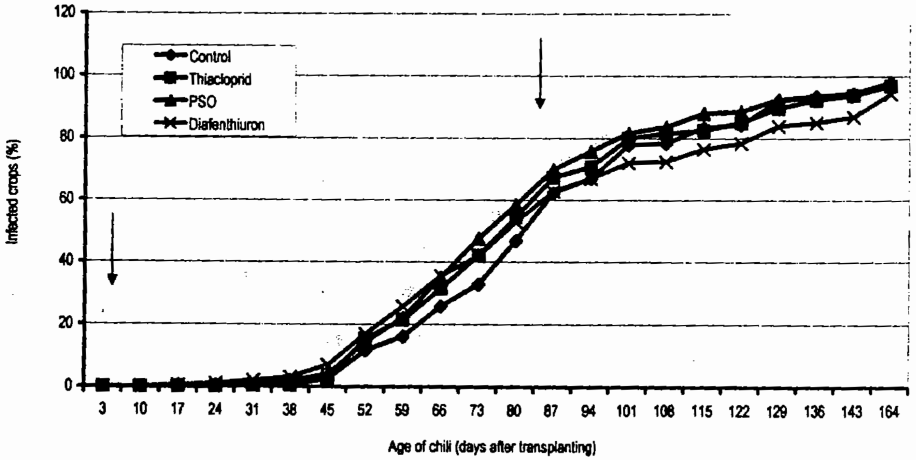

Fig. 3. The seasonal development of disease in the field plots treated with different insecticides. The first arrow showed the first insecticides application, while the second arrow represented the last insecticides application. Insecticides were applied weekly.

Disease incidence. Changes in average numbers of *B. tabaci* adults in all plots were followed by similar changes in the disease incidence. An increase in the number of adult *B. tabaci* (Fig. 1) caused a sharp increase in the severity of disease incidence in the fields (Fig. 3). At the end of the season as the number of adults caught decreased (Fig. 1), the disease incidence increased at lower rate (Fig. 3). Eventually, significant differences in percentage of infected plants among treated and untreated plots were noticed. The severity of the disease in all treatments was considerably very high (>94%, Fig. 3).

Yield. Application of insecticides did not provide effective control of the disease, even though the insecticides did decrease the population of the vector. Although chili crops that were sprayed with thiacloprid gained higher yield than other plots (Fig. 4), the yield was so much reduced from its potential yield (±1.3 kg/plant; Oriental Seed Indonesia, pers. comm.) because of the disease infection.

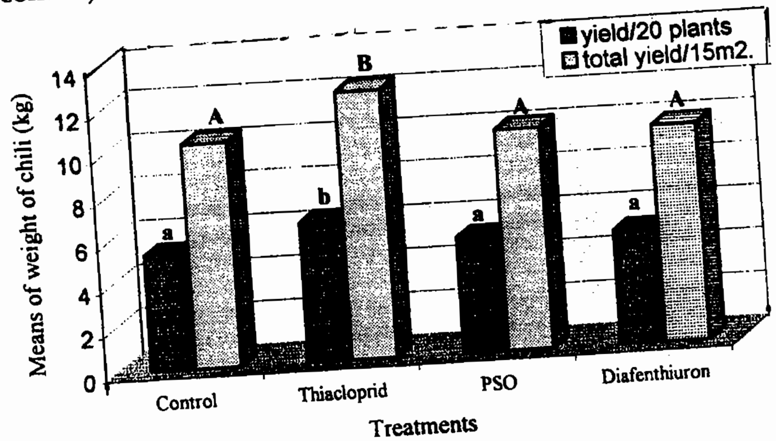

Fig. 4. The yield of chilli produced from plots treated with different insecticides. Means followed by similar lower case (yield/20 plants) or upper case letter (total yield/15m²) are not significantly different at á =5% based on Fisher's protected LSD test.

Confirmation of the disease. PCR analysis was able to amplify 2.8 kb of geminivirus viral gene (Fig. 5).

Fig. 5. Electroforesis of PCR product amplified using universal primers of Begomovirus: DNA marker (line 1), positive control of Tomato Yellow Leaf Curl Virus (line 2), sample of healthy chili (line 3), and sample of infected chili (line 4).

The results showed that insecticides treatments were only able to decrease *B. tabaci* population only for certain time. Plots treated with diafenthiuron had lower adult densities compared to the control plots. Under field condition, this compound will be phytochemically converted to a carbomiide derivative in the presence of sunlight that make its insecticidal activity greater than the parents compound (Steinemann *et al.* 1990 *cit* Palumbo *et al.*, 2001b). This also make this compound has a high toxicity to adult if it is applied as a foliar insecticide. Contact with diafenthiuron might happen when they feed on plant sap or rest on plants.

PSO has been used traditionally to control small, relatively immobile insect by suffocation (Rae *et al.*, 1996), the most likely mode of action of oil against the nymphs. PSO acts on adults as repellence (Stansly *et al.*, 2000). High number of adults was observed in PSO plots and it was similar to the control plots. This might be caused by the fact that PSO only repels the adults instead of killing them. Therefore, the adults still can be found alive in the plants. This phenomenom was previously reported by Stansly *et al.* (2000), indicated that PSO was not effective in suppressing adults of *B. tabaci* on tomato.

Application of thiacloprid could maintain the adults densities below the densities in the control plots during the population peak. Thiacloprid, the most recent nicotinoid compound, has been studied to be a good persistent translaminar activity as a foliar spray and shown residual efficacy against *B. tabaci*. It provided a good adult knockdown and reduction in egg deposition (Palumbo *et al.*, 2001a). Efficacy of this compound against *B. tabaci* also reported on melon (Palumbo *et al.*, 2001b).

Suppression of adults or nymphs densities did not reduce the severity of the disease. Disease spreads rapidly in the field along with the increasing number of adults. They may feed only a short time on a plant then move to neighboring plants. If this hypothesis is accepted, it accounted significantly for the fact that all plots got high infection (± 94%) of begomovirus though insecticides had been applied periodically. Infection of the virus in the early developmental stage will result in little to no production (Brown, 1998). In this case, high infection did not occur in the early season but it occurred at the nearly flowering stage so that chili plants could produce fruits. However, the yield was so much reduced from its potential yield.

Universal primers of begomovirus that were used were able to amplify 2.8 kb viral genom from infected chili. The PCR detection showed that the disease on chili crops in experimental field was viral disease caused by a member of virus belong to the group of begomoviruses.

CONCLUSION

Weekly application of the insecticides started at the beginning of the season until a week before harvest did not provide complete protection to the yield. The disease incidence increased progressively during the season, eventhough the insecticides application did decrease significantly the population of adults and nymphs of *B. tabaci* during the peak season (66-101 DAT). The disease was positively identified as a virus disease caused by begomovirus, similar to Tomato Yellow Leaf Curl Virus.

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